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IN THE

Supreme Court of the United States

RICHARD E. GLOSSIP, *et al.*,

Petitioners,

v.

KEVIN J. GROSS,

Respondent.

ON WRIT OF CERTIORARI TO THE UNITED STATES COURT OF APPEALS FOR THE TENTH CIRCUIT

BRIEF OF SIXTEEN PROFESSORS OF PHARMACOLOGY AS *AMICI CURIAE* IN SUPPORT OF NEITHER PARTY

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STATEMENT OF INTEREST OF *AMICI CURIAE*

Amici curiae, each of whom is listed below, are professors of pharmacology at universities in the United States.[1] *Amici curiae* respectfully submit this brief to provide a pharmacological perspective on the physiologic effect of midazolam hydrochloride ("midazolam"). Midazolam is a sedative in the benzodiazepine class of drugs that the State of Oklahoma decided to use in early 2014 as a substitute for sodium thiopental ("thiopental") and pentobarbital as the first drug in the State's three-drug lethal injection protocol. *Amici curiae* have no interest in any party to this litigation, nor any stake in the outcome of this case.[2]

Edward Bilsky, Ph.D. is the Vice President for Research and Scholarship and a Professor of Pharmacology at the University of New England in Biddeford, ME. At the University of New England, Dr. Bilsky is also the Founding Director of the Center for Excellence in the Neurosciences. Dr. Bilsky received a B.S. in Physics from Rensselaer Polytechnic Institute in Troy, NY; a M.S. in Behavioral Neuroscience from Rensselaer

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[1] No counsel for a party authored this brief in whole or in part, and no such counsel or party made a monetary contribution intended to fund the preparation or submission of this brief. No person other than the *amici*, or their counsel, made a monetary contribution intended to fund its preparation or submission. The parties have filed blanket waivers with the Court consenting to the submission of all *amicus* briefs.

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Letters from the parties consenting to the filing of *amicus curiae* briefs, in support of either party or neither party, have been filed with the Clerk of the Court.

SUMMARY OF ARGUMENT

The first drug in the State of Oklahoma's three-drug lethal injection protocol is intended to ensure an inmate is in a "deep, comalike unconsciousness" prior to the injection of a paralytic agent to stop respiration, and a drug that induces cardiac arrest. *Warner v. Gross*, 776 F.3d 721, 724-25 (10th Cir. 2015), *cert. granted*, 135 S. Ct. 1173 (2015). Since early 2014, the State of Oklahoma has used midazolam as the first drug in this sequence. From a pharmacological perspective, however, midazolam is not appropriate for its intended purpose.

There is overwhelming scientific consensus, including among pharmacologists, that midazolam is incapable of inducing a "deep, comalike unconsciousness," *id.* The biochemical interaction by which midazolam produces its pharmacological effect on the human body (i.e., its mechanism of action) requires co-binding of a neurotransmitter, known as γ-aminobutyric acid ("GABA"), and midazolam to depress the activity of the central nervous system ("CNS"). But the depth of that depression is limited, and even an excessive dose of midazolam will not result in unconsciousness.

ARGUMENT

I. Background

Before the Director of Oklahoma's Department of Corrections selected midazolam for use as the first drug in the three-drug lethal injection protocol in early 2014, the State of Oklahoma used drugs in the barbiturate class known to "induce[] a deep, comalike unconsciousness when given in the amounts used for lethal injection." *Id.* at 724-25 (citing *Baze v. Rees*, 553 U.S. 35, 44 (2008)). Until approximately 2010, the State of Oklahoma used thiopental, a member of the barbiturate drug class, as the first drug. *Id.* at 725. Later, it substituted an alternative drug in the same barbiturate class, pentobarbital, as the first drug in the protocol. *Id.* But in 2014, the State of Oklahoma elected to use midazolam in its place. *Id.*

Midazolam is not an appropriate substitute for thiopental or pentobarbital as the first drug in the State of Oklahoma's three-drug lethal injection protocol. Increasing doses of barbiturates, including thiopental and pentobarbital, will induce sedation, then sleepiness, then anesthesia, then coma, and finally death. Midazolam, however, is not a member of the barbiturate drug class. It is a benzodiazepine, a separate class of drugs that does not exert its pharmacological effect in the same manner as barbiturates. This pharmacological distinction explains why midazolam cannot induce unconsciousness at any dose, and why it is not an appropriate substitute for either thiopental or pentobarbital as the first drug in a three-drug lethal injection protocol.

II. Midazolam Is a Fast-Acting Benzodiazepine that Produces Reliable Hypnosis, Amnesia, and Anti-anxiety Effects

Midazolam was first synthesized in 1976 and belongs to a class of drugs called benzodiazepines. J.G. Reves *et al.*, *Midazolam: Pharmacology and Uses*, 62 Anesthesiology 310, 310 (1985). Benzodiazepines are CNS depressants that reliably provide sedative, hypnotic[3], muscle relaxant, anxiety inhibitory, and anticonvulsant effects. Dennis S. Charney *et al.*, *Hypnotics and Sedatives, in Goodman & Gilman's The Pharmacological Basis of Therapeutics* 401, 410-11 (James F. Shanahan *et al.* eds., 11th ed. 2006); Reves at 311-12. In addition to midazolam, the benzodiazepine family also includes the active pharmaceutical ingredients in XANAX® (alprazolam), KLONOPIN® (clonazepam), VALIUM® (diazepam), and ATIVAN® (lorazepam), among others. Charney at 410-11.

Structurally speaking, midazolam differs from other benzodiazepines in that it has a fused imidazole ring, which confers unique physiochemical properties, including basicity (as opposed to acidity), stability in aqueous solution, and rapid metabolism in the body. Reves at 310. At physiologic pH (i.e., pH of the blood), midazolam becomes highly lipophilic (i.e., able to dissolve in fats), giving rise to

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[3] Hypnotic drugs "produce[] drowsiness and facilitate[] the onset and maintenance of a state of sleep that resembles natural sleep in its electroencephalographic characteristics and from which the recipient can be aroused easily." Charney at 401.

various clinical consequences, including rapid entry of midazolam into brain tissue and very rapid onset of activity following intravenous administration. *Id.* at 315. Although it is fast acting, midazolam also has a short duration of activity due to its very high metabolic clearance and rapid rate of elimination from the body. *Id.* Termination of the clinical effect following a single dose of midazolam is also rapid. *Id.*

These properties make midazolam an attractive choice as a pre-medication given prior to the induction of anesthesia. *Id.* at 317-18. But while midazolam produces sleep and amnesia, with a short duration of activity, it cannot be used to render a person unconscious or to maintain general anesthesia. *Id.*

III. Midazolam Is Incapable of Rendering an Individual Unconscious

Midazolam's mechanism of action differs from barbiturates. This critical difference demonstrates why midazolam cannot induce unconsciousness, and why it is an improper substitute for thiopental or pentobarbital as the first drug in the State of Oklahoma's three-drug lethal injection protocol.

A. Background on Neurotransmission and Neuronal Inhibition

To understand how midazolam works on the body—and ultimately to convey why midazolam is incapable of "render[ing] an inmate unconscious prior to the injection of the second and third drugs," *Warner*, 776 F.3d at 725—requires some background

on neurotransmission, the process by which neurons (i.e., nerve cells) receive, conduct, and transmit signals.

Neurotransmission is at the heart of every action in our daily lives, taking a stimulus, converting it into a nerve impulse, and resulting in some action or inaction. When we touch a hot burner on a stove, we immediately pull our hand away. In the home, this is a clumsy kitchen mishap. But in the classroom, this is a classic lesson in neurotransmission. At the biochemical level, touching the hot burner stimulates heat and pain receptors on the hand, which send nerve impulses to the spinal cord. Those nerve impulses are then transferred to neurons that are ultimately connected to the muscles in the arm and hand that are stimulated to contract to pull the hand away. This cascade of events is the result of a series of electrochemical reactions mediated by neurotransmitters. Neurotransmitters are chemicals that bind to receptors, causing ion channels to open or close, thus exciting or inhibiting individual neurons.

GABA is the major inhibitory neurotransmitter in the human body. *E.g.*, Bruce Alberts *et al.*, *Membrane Transport, in Essential Cell Biology* 371, 400-01 (1998); Charney at 405. When inhibitory neurons of the brain release GABA onto other brain neurons, GABA binds to GABA-specific receptors. Alberts at 400-01. This binding causes chloride ion channels to open on the recipient neurons. *Id.* The influx of chloride ions through the channel causes those neurons to become more

quiescent, to decrease in electrical activity, and to decrease the likelihood of neuronal firing. *Id.* at 401. The result is neuronal inhibition and ultimately CNS depression. *Id.*

By way of example, seizures result from a failure of inhibitory neurotransmission, and there is substantial evidence linking epilepsy with dysfunction of GABA-specific neuronal inhibition. L. John Greenfield Jr., *Molecular Mechanisms of Antiseizure Drug Activity at $GABA_A$ Receptors*, 22 Seizure 589, 589 (2013). Drugs that block or dampen GABA's effect on binding to the GABA receptor are called "antagonists." *Id.* By preventing or decreasing the likelihood of chloride ion influx through the ion channel, these antagonists can induce seizures. *Id.* Conversely, drugs that activate the GABA receptor are called "agonists," and by increasing the frequency of ion channel opening, or increasing the opening time of the ion channel to longer durations, agonists work to sustain neuronal inhibition and control seizures. *Id.* at 590-92. Even outside of the context of seizures, pharmaceutically promoting neuronal inhibition requires agonists that enhance the chloride ion channel permeability of GABA receptors. *E.g.*, Alberts at 401.

B. Midazolam Promotes CNS Depression Only in the Presence of GABA by Increasing the Frequency of Ion Channel Opening

Once in the body, midazolam, like all benzodiazepines, acts by promoting the binding of the neurotransmitter GABA to the $GABA_A$ receptor. Charney at 402; Reves at 311-12; Greenfield at 591.

$GABA_A$ receptors, a sub-type of GABA receptors, are ion channels with multiple binding sites that can be opened by GABA, but that also can be modulated by drugs, including benzodiazepines. Werner Sieghart, *Allosteric Modulation of $GABA_A$ Receptors via Multiple Drug-Binding Sites*, 72 Advances in Pharmacology 53, 54-55 (2015).

When midazolam alone binds to the $GABA_A$ receptor, there is no inhibitory neuronal consequence. Erwin Sigel & Michael E. Steinmann, *Structure, Function, and Modulation of $GABA_A$ Receptors*, 287 Journal of Biological Chemistry 40224, 40227 (2012); Charlotte D'Hulst *et al.*, *The Complexity of the $GABA_A$ Receptor Shapes Unique Pharmacological Profiles*, 14 Drug Discovery Today 866, 872 (2009); Charney at 405. In other words, the binding of midazolam alone to the $GABA_A$ receptor does not open the ion channel to allow for chloride ion influx into the neuron. Rather, midazolam requires the additional co-binding of GABA to the $GABA_A$ receptor to exert an inhibitory effect. D'Hulst at 872; Charney at 405-06; Werner Sieghart *et al.*, *A Novel $GABA_A$ Receptor Pharmacology: Drugs Interfacing with the $\alpha^+\beta^-$ Interface*, 166 British Journal of Pharmacology 476, 476-77 (2012). This requires coordination, as neuronal inhibition by midazolam therefore requires GABA to be released by inhibitory neurons and available to bind to the $GABA_A$ receptor at the same time that the drug is available to bind. D'Hulst at 872; Charney at 405-06; Sieghart (2012) at 477.

Midazolam and GABA bind to the $GABA_A$ receptor at distinct sites. Brett A. Cromer *et al.*,

Anxiety over $GABA_A$ Receptor Structure Relieved by AChBP, 27 Trends in Biochemical Sciences 280, 280 (2002); M. Ernst *et al.*, *Comparative Modeling of $GABA_A$ Receptors: Limits, Insights, Future Developments*, 119 Neuroscience 933, 938-39 (2003). When midazolam binds to the benzodiazepine site on a $GABA_A$ receptor, this leads to a conformational change of the receptor itself. Sigel & Steinmann at 40227. The conformational change increases the affinity of the GABA neurotransmitter to its own $GABA_A$ receptor binding site. *Id.*; Charney at 406. The increased affinity of GABA for the $GABA_A$ receptor binding site means that in the presence of bound midazolam, even sub-maximal amounts of GABA will open the $GABA_A$ ion channel. Sigel & Steinmann at 40227; Charney at 406.

The result of co-application of midazolam and GABA to the $GABA_A$ receptor is an increase in the frequency of chloride ion channel opening. Sigel & Steinmann at 40228; Robert E. Study & Jeffrey L. Barker, *Diazepam and (–)-Pentobarbital: Fluctuation Analysis Reveals Different Mechanisms for Potentiation of γ-aminobutyric Acid Responses in Cultured Central Neurons*, 78 Proceedings of the National Academy of Sciences USA 7180, 7183 (1981); Carl J. Rogers *et al.*, *Benzodiazepine and β-carboline Regulation of Single $GABA_A$ Receptor Channels of Mouse Spinal Neurones in Culture*, 475 Journal of Physiology 69, 69, 72-73, 77, 79 (1994); Charney at 406. The resultant influx of chloride ions into the neuron suppresses neuronal firing, and ultimately produces the hallmark sedative and hypnotic effects of the drug. *E.g.*, Alberts at 400-01; D'Hulst at 866.

By way of analogy, the $GABA_A$ receptor can be thought of as a keyed gate through which chloride ions can enter a neuron. GABA and midazolam are keys to that gate. The GABA key alone opens the gate, allowing for chloride ion influx. The midazolam key alone does not open the gate, but simultaneous use of the GABA and midazolam keys results in increased frequency of gate opening, as compared to the frequency of gate opening using the GABA key alone.

C. Barbiturates Depress the CNS by Increasing the Duration of Ion Channel Opening Even in the Absence of GABA

Barbiturates, including thiopental and pentobarbital, also act on the $GABA_A$ receptor. *E.g.*, Richard W. Olsen & Adele M. Snowman, *Chlorine-Dependent Enhancement by Barbiturates of γ-aminobutyric Acid Receptor Binding*, 2 The Journal of Neuroscience 1812, 1812 (1982); Greenfield at 589; Charney at 414-16. The binding site for barbiturates is located in a different region of the $GABA_A$ receptor, distinct from the binding site for benzodiazepines. Ismar Newton Cestari *et al.*, *The Agonistic Action of Pentobarbital on $GABA_A$ β-subunit Homomeric Receptors*, 7 NeuroReport 943, 946 (1996); Charney at 414. While benzodiazepines enhance GABA inhibition by modulating the frequency of ion channel opening, barbiturates enhance GABA inhibition by increasing the time the ion channel remains open. Study & Barker at 7182-83; Charney at 406, 414.

Unlike benzodiazepines, which require the presence of GABA to produce a pharmacologic effect,

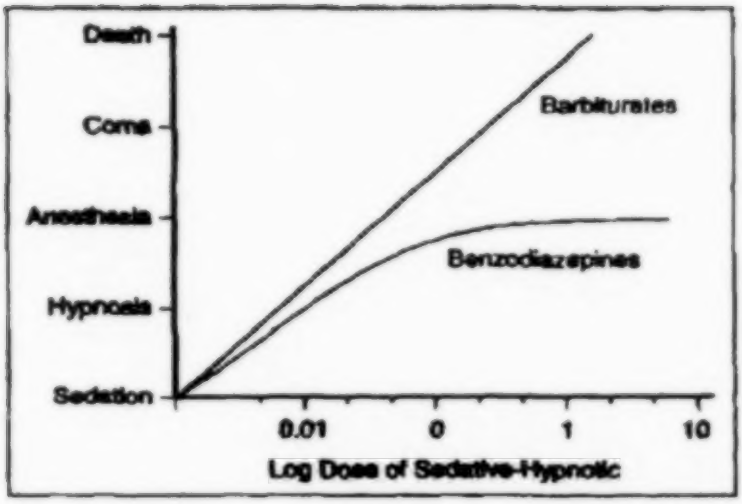
barbiturates confer neuronal inhibition even in the absence of GABA. David A. Mathers & Jeffrey L. Barker, *(–)-Pentobarbital Opens Ion Channels of Long Duration in Cultured Mouse Spinal Neurons*, 209 Science 507, 507-08 (1980); Meyer B. Jackson *et al.*, *Single Channel Currents Activated by γ-aminobutyric Acid, Muscimol, and (–)-Pentobarbital in Cultured Mouse Spinal Neurons*, 2 The Journal of Neuroscience 889, 890-93 (1982); Charney at 406. Therefore, the binding of barbiturates to the $GABA_A$ receptor opens the chloride ion channel in the absence of GABA and keeps the channel open longer than benzodiazepines. Robert L. Macdonald *et al.*, *Barbiturate Regulation of Kinetic Properties of the $GABA_A$ Receptor Channel of Mouse Spinal Neurones in Culture*, 417 Journal of Physiology 483, 491-96 (1989); Feyza Sancar & Cynthia Czajkowski, *Allosteric Modulators Induce Distinct Movements at the GABA-binding Site Interface of the GABA-A Receptor*, 60 Neuropharmacology 520, 527 (2011); Charney at 406, 414. In other words, even in the absence of GABA, barbiturates can increase the influx of chloride ions into the neuron to completely shut down the activity of the neuron.

D. Because Midazolam Requires Co-Binding of GABA and the Amount of GABA is Limited, Midazolam Cannot Overcome Sub-Anesthetic Levels To Provide General Anesthesia

As noted above, when a drug activates its receptor, it is called an agonist. A drug that activates its receptor to produce a maximal effect is called a full agonist; one that cannot produce the maximal

effect is called a partial agonist. Because benzodiazepines facilitate GABA actions, they are considered agonists. *E.g.*, Trevor J. Anthony & Walter L. Way, *Sedative-Hypnotic Drugs, in Basic & Clinical Pharmacology* 379, 379 (Bertram G. Katzung *et al.* eds., 12th ed. 2012); D'Hulst at 872. Yet because benzodiazepines require co-activation with GABA to produce a pharmacologic effect, and the amount of GABA is limited, they can produce only a partial agonist effect. George M. Brenner & Craig W. Stevens, *Sedative-Hypnotic and Anxiolytic Drugs, in Pharmacology* 186, 192 (Fig. 19-3) (4th ed. 2013) Barbiturates, on the other hand, act to inhibit neurons even in the absence of GABA and produce full agonist effects. *Id.* Thus, midazolam is only a partial agonist, and the depth of its inhibitory effect has limits.

This difference between full agonist and partial agonist explains why barbiturates like thiopental and pentobarbital can induce a "deep, comalike unconsciousness when given in the amounts used for lethal injection," *Warner*, 776 F.3d at 724 (citing *Baze*, 553 U.S. at 44), and why midazolam is incapable of "render[ing] an inmate unconscious prior to the injection of the second and third drugs," *id.* at 725. This difference, referred to as the "ceiling effect" of the benzodiazepine response, is illustrated in the figure below.



Brenner & Stevens at 192 (Fig. 19-3). As depicted above, greater doses of benzodiazepines will not produce greater pharmacological effects, while greater doses of barbiturates will produce greater pharmacological effects. *See id.* This is for two reasons. First, benzodiazepines require the presence of GABA to exert a pharmacological effect and barbiturates do not. *E.g.*, Charney at 405-06. Yet GABA is present in limited supply in the CNS, and the limited quantity of GABA restricts the pharmacological effect of benzodiazepines. *See, e.g.*, *id.* at 406. Second, benzodiazepines work differently. Specifically, benzodiazepines merely increase the frequency of ion channel opening when GABA is also present. Sigel & Steinmann at 40228; Study & Barker at 7183; Rogers at 69, 72-73, 77, 79; Charney at 406. Unlike barbiturates, benzodiazepines do not affect the duration of ion channel opening. *E.g.*, Charney at 406.

With reference to the increasing CNS effects on the vertical axis in the figure above, as the dose of benzodiazepine increases, the benzodiazepine curve

plateaus, reaching a "ceiling" before general anesthesia can be obtained. *See* Brenner & Stevens at 192 (Fig. 19-3). Therefore, the response to benzodiazepines cannot be further enhanced to unconsciousness and beyond by increasing the dose. *See id.* The fact that midazolam does not ever achieve general anesthesia is consistent with and explains the clinical uses of the drug. In fact, it is widely recognized in the scientific and medical community that midazolam alone cannot be used to maintain adequate anesthesia for surgery. *E.g.*, Reves at 318; Charney at 404 ("The clinical literature often refers to the 'anesthetic' effects and uses of certain benzodiazepines, but the drugs do not cause a true general anesthesia because awareness usually persists, and relaxation sufficient to allow surgery cannot be achieved."). By contrast, barbiturates do not exhibit this ceiling effect. *See* Brenner & Stevens at 192 (Fig. 19-3). Accordingly, increasing doses of barbiturates reliably produce anesthesia, coma, and death. *See id.*

In sum, midazolam's mechanism of action makes it unsuitable as the first drug in the State of Oklahoma's three-drug lethal injection protocol because it is incapable of inducing unconsciousness.

CONCLUSION

From a pharmacological perspective, midazolam is not interchangeable with barbiturates like thiopental or pentobarbital. Midazolam is incapable of rendering an inmate unconscious prior to the injection of the second and third drugs in the State of Oklahoma's lethal injection protocol. Therefore, midazolam is not appropriate for its

intended purpose as the first drug in the State of Oklahoma's three-drug lethal injection protocol.

Respectfully Submitted,

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